

Claims

1. A device for analyzing sample arrays deposited on a substrate surface, the device comprising:

a stage for holding the substrate, said stage movable along a first axis of said surface;

at least one laser producing an illumination beam;

focus optics in the path of the illumination light, said illumination focus optics focusing said illumination beam into a beam spot;

a beam scanner for scanning said beam spot along a second axis of said surface;

an objective lens, which gathers a percentage of light emitted from samples on said substrate and transmits collected light along an optical axis of said illumination beam to a light detector;

a beam directing optic positioned between said laser and said objective lens said beam directing optic including an inner light directing element that directs the illumination beam to the objective lens and an outer light directing element disposed as an annulus about the inner light directing element, said element directing collected emitted light at an angle relative to the optical axis of the illumination beam to detection optics;

a spatial filter, acting in conjunction with the beam spot and objective lens to confine detection to a limited depth of field, said depth of field in a defined spatial relation to said substrate surface; and

a light detection optics positioned to detect light which passes through said spatial filter.

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2. The device of claim 1, further comprising:

a beam splitting optics, positioned between said spatial filter and said light detector, wherein said beam splitting optics splits collected light into two spectral components that are separately detected by the light detection optics.

3. The device of claim 2, further comprising:

a computer that characterized said collected emitted light by comparing ratio of measured intensity of each spectral component.

4. The device of claim 1, further comprising:

at least one optical filter positioned between said laser and said focus optics such that said illumination beam passes through the filter.

5. The device of claim 1, wherein the at least one laser includes a first laser and a second laser and the device further comprises beam combining optics to combine beams produced by said first and second laser to produce the illumination beam.

6. The device of claim 1, wherein said beam scanner is one of a group comprising a galvanometer, a Bragg cell, a resonant scanner and a rotating polygonal mirror, said device moving said beam spot in a line scan.

7. The device of claim 1, further comprising:

beam spot focusing optics, said optics detecting when the beam spot is focused onto said substrate surface.

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8. The device of claim 1, wherein said inner light directing optic is positioned proximate to a waist of the illumination beam.

9. A method to optically analyze discrete, optically detectable targets, the method comprising:

a) directing a focused beam spot of an illumination beam using a light directing optic onto the targets;

b) collecting emitted light with a wide angle light collector and transmitting said light as a retrobeam along pathway of said illumination beam using said light directing optic to a spatial filter;

c) focusing collected light through said spatial filter, whereby light from outside a focal depth is excluded by said spatial filter from reaching light detectors thereby limiting the depth of field to 1-100 μm ;

d) detecting an intensity of at least two spectral components; and

e) analyzing said detected emission intensity such that wavelength profiles may be detected, wherein the number of wavelength profiles that may be detected is greater than the number of spectral components detected, wherein said light directing optic includes an inner illumination beam directing optic which directs the illumination beam to the sample.

10. The method of claim 9, wherein analyzing said detected emission is performed by calculating a ratio of said spectral components.

11. The method of claim 9, wherein the step of directing a focused beam spot includes focusing a beam waist into a target layer.

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12. The method of claim 11, wherein focusing a beam waist to a target layer includes using specular reflection to locate a reference surface, said surface having a spatially defined relation to said layer.

13. The method of claim 9, wherein step c includes collecting light with an objective lens.

14. The method of claim 9, wherein step b is performed by optically scanning the beam spot in a first direction on the surface of said substrate and mechanically translating said substrate in a tangent direction.

15. A system for detecting discrete fluorescent targets, the system comprising:

at least one laser producing an illumination beam;

illumination focus optics in the path of the illumination beam, said illumination focus optics focusing said illumination beam into a beam spot;

a light directing optic positioned between said focus optics and said beam scanner, said light directing optic including a central light directing optic surrounded by an outer light directing annulus, wherein the central light directing optic has a width matched to a waist of focused illumination beam and said central optic directs the beam spot to said layer;

an objective lens, which focuses the beam spot onto the sample, thereby exciting fluorescent emission, said objective further collecting said fluorescent emission and directing as a retrobeam said emission light to said outer light detecting annulus, wherein said outer annulus directs collected fluorescent light onto detection optics;

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a spatial filter that confines detection to a limited depth of field;

a light detector positioned to detect light which passes through said spatial filter; and

a computer receiving signals from said detector, said computer analyzing said signals to recognize discrete targets.

16. The system of claim 15, wherein the light directing optic is a dot mirror with a transparent annulus, wherein the dot mirror directs the illumination light to the sample and the transparent annulus allows collected fluorescent light to pass onto the detection optics.

17. The system of claim 15, wherein the light directing optic is a mirror with a beam pass through hole located proximate to a center of the mirror, wherein the mirror is positioned such that the illumination beam may pass through the hole while the collected emission light is reflected by the mirror onto detection optics.

18. The system of claim 17, wherein the hole is positioned proximate to a waist of the illumination beam.

19. The system of claim 15, further comprising,
at least one laser line filter positioned between said laser and said focus optics such that said illumination beam passes through the laser line filter.

20. The system of claim 15, further comprising shutters placed between the laser and the illumination focus optics.

21. The system of claim 15, further comprising a beam scanner that scans the beam spot through a layer in a sample container holding the fluorescent targets.

22. The system of claim 21, wherein said beam scanner includes a device selected from the group comprising a galvanometer, a Bragg cell, a resonant scanner and a rotating polygonal mirror, said device moving said beam spot in a line scan.

23. The system of claim 22, wherein the beam scanner further comprises a stepper motor for translating the sample in a direction tangent to said line scan.

24. The device of claim 15, further comprising:
a focusing system for targeting the beam spot onto the layer on the sample.

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